IV. THE INVENTION DISCLOSED AND CLAIMED HEREIN

The essence of the present invention is the discovery that carbohydrate antigens indigenous to either Gram-stain-negative or Gram-stain-positive bacteria, if separated from cultures of the bacteria to which they are indigenous and rendered essentially protein-free, i.e., to the point that they contain not more than about 10% protein and preferably less, are exceptionally efficient, when coupled to a chromatographic affinity column, in imparting antigen-specificity to crude antibodies (raised either to the bacteria or to an Ig G cut of the crude antigen) when these antibodies are passed over the column. The resulting purified antibodies are superior agents of exceptional sensitivity and specificity toward the crude carbohydrate antigen embodiments of the same carbohydrate antigen that has been rendered essentially protein-free.

This discovery has afforded an outstanding basis for the development of rapid disposable immunochromatographic ("ICT") diagnostic tests that identify the crude antigen of the bacteria in human fluids and can be run, without special equipment, on a sample of human bodily fluid, even by persons lacking laboratory training and at virtually any location. These tests can especially be expeditiously be run on readily available human body fluid samples, such as urine (which is noninvasively obtainable), blood or serum, spinal fluid, sputum, middle ear fluid, etc. The ability to identify the bacteria quickly and associate them with a patient's clinical symptoms and signs enables rapid diagnosis of the patient's disease state and rapid, accurate medication of the patient.

The tests are of particular value because many of the diseases caused by bacteria have, even in the recent past, been considered extremely difficult to diagnose without a bacterial culture. The disease states, in turn, have been hard to conquer because patients could not be adequately (or, even in some instances, appropriately) medicated during the period of some days necessary for the obtainment of an adequate culture.

V. RESPONSE TO PARAGRAPH 6 OF THE ACTION --"ENABLEMENT"

The enablement rejection of the action is unclear and unprecedented. There is not now and never has been a requirement, in the statute, the rules, the MPEP or any case of which Applicants' counsel is aware that requires a method claim to be conducted in one continuous operation from start to finish.

In the particular overall methodology disclosed in *this* case, it is clear that culturing bacteria, separating a carbohydrate antigen therefrom and treating that antigen to reduce its protein content as far as practical limits permit, to not more than and usually somewhat less than 10%, coupling that antigen to a chromatographic column and then passing crude, conventionally obtained antibodies thereover to purify them, are all steps that must be conducted in a properly outfitted immunology laboratory by technically qualified, well-trained immunology professionals and laboratory assistants. The design of an immunochromatographic test also requires technically qualified, trained immunology professionals. The manufacture of these tests, however, can be effected by persons without knowledge of immunology beyond the manufacturing training given by the manufacturer on-site. The preferred immunochromatographic tests themselves may be performed by anyone who is able to read and comprehend the directions with which the ICT test strips are packaged prior to shipping.

The purified antibodies obtained by the chromatographic affinity purification of crude antibodies by passing them over the essentially protein-free carbohydrate antigen on the chromatographic affinity column have enhanced sensitivity and specificity for the same crude carbohydrate antigen, an embodiment of which in "essentially protein free" form is coupled to the matrix on the column. The purified antibodies can thereafter be appropriately disposed on an immunochromatographic strip in a way that permits a rapid test to be carried out in virtually any environment, by almost anyone who can read how to perform the test. The sample preferred is human urine because of its ready availability and the fact that it can be collected noninvasively of the patient; the sample can, however, be another human patient fluid such as blood, serum, saliva, sputum or any other human fluid suspected of carrying bacteria and bacteria fragments causative of infection, such as spinal tap fluid, middle ear fluid, etc.

The test strips are prepared in a manufacturing milieu for placement in a housing, packaging and distribution to hospitals, clinics, physician's offices and like places which are the major users of the products.

Because the carbohydrate antigens are typically bifunctional, the ICT strips are prepared by placing a movable deposit of labelled, purified antibodies near the point of introduction of the sample, so that they will be picked up by, and flow with, the liquid sample to an immovably deposited, fixed "capture line" of unlabelled purified antibodies placed near the opposite end of the strip. The essence of the test reaction that occurs, if the sample contains the target antigen, is in two steps. First, labelled purified antibodies conjugate to target antigen in the sample as the labelled antibodies and sample flow together toward the capture line. Second, labelled antibody-antigen conjugates react at the capture line with the

immobilized antibodies deposited there, to form "sandwiches" of labelled antibody-antigen-immobilized antibody.

The essence of the invention lies in the purified antibodies which exhibit greatly enhanced specificity and sensitivity toward the target carbohydrate antigen in the sample and accordingly give test results that, when combined with clinical symptoms of the patient, enable rapid diagnoses of bacterially-caused disease states and prompt prescription of appropriate medication.

The Examiner presents no rationale for the suggestion that unless all of the steps needed to produce the test result are combined into a single hybrid method example, those of ordinary skill in immunology would be incapable of reproducing the methodology and attaining the stated result. The suggestion itself is unprecedented and has no support in the patent statutes, the Rules of Practice, the M.P.E.P. or any case that applicants' counsel has been able to locate. Furthermore, it flies in the face of common experience to suggest that greater clarity is achieved if methodology is condensed into a single complicated procedure instead of being described step by step.

The same methodology can, as applicants have discovered, be utilized with either gram-negative or gram-positive bacteria. The specification clearly teaches that the methodology applies to both gram-negative and gram-positive bacteria. Applicants know of no bacteria species, or serogroup of a species, that simultaneously is *both* gram-negative and gram-positive and understands that the Gram stain is used to differentiate two types of bacteria on the basis of their reaction to the stain. If the sentence bridging pages 5 and 6 of the Action is intended to suggest the existence of a type of bacteria that simultaneously exhibits characteristics of both

gram-stain positivity and gram stain negativity, the sentence is not understood.

Applicants have presented herewith the declaration of William J. Palin, who has worked in the field of immunology for some 35 years. Dr. Palin is currently employed as Vice-President of Research by applicants' assignee, Binax, Inc, which he joined in 2001. He is intimately familiar with the immunology field, and is well-qualified to attest to the vast amounts of information relating to that field that are today readily available to those of ordinary skill in the art. His declaration refutes the overall conclusion of the office action that one of ordinary skill in immunology could not practice the invention as described in the application (including in the material incorporated by reference). It also rebuts various specific statements in the Office Action.

Other parts of this enablement rejection appear to criticize the methodology of the application simply because it has been ascertained that carbohydrate antigens characteristic of a particular bacteria species, or a particular serogroup of a species, in general, *can* be subjected to this methodology. It should be understood that many bacteria have long been known to possess more than one such characteristic carbohydrate antigen--and that, as the specification teaches and Dr. Palin's declaration affirms--one desiring to devise a useful test according to this invention will normally consult the published scientific literature to determine (a) what has been previously determined with respect to the carbohydrate antigens of the bacteria species, or serogroup of a species, for which a rapid test is needed or desired and (b) how each such carbohydrate antigen has previously been separated from a bacterial culture. From this study, the carbohydrate antigen deemed most practical to target is selected and a method for obtaining it in essentially protein-free form (i.e. not more, and preferably less, than 10% by weight of

protein) is either adopted from the literature or is devised, using public knowledge readily available to and useable by persons that have training in immunology. Much has been published about methods for separating carbohydrate antigens from their host bacteria and much has also been published about how to minimize the protein content of such a carbohydrate antigen, by treatments incorporated in the separation procedure and in other ways, such as after-treatments. The steps of coupling the antigen to a chromatographic affinity column, followed by passing crude antibodies raised to either of (a) the bacteria species, or serogroup of a species, from which the purified carbohydrate antigen was obtained or (b) an Ig G cut of the crude carbohydrate antigen *per se*, are steps that can readily be performed by those trained in immunology using the information given in this application (including information incorporated by reference) and other information readily available to them from literature and from suppliers, e.g., of affinity matrices for chromatographic affinity columns and suppliers of various reagents. The design of a rapid ICT test wherein the resulting purified antibodies are utilized to detect the crude antigen in a sample of fluid taken from a human subject such as blood, urine, sputum, etc. can be achieved readily by a person trained in immunology by following the directions given herein (including those incorporated by reference) and taking advantage, as needed, of information available in a wide range of publicly available literature. The actual performance of the test can be done by almost anyone. Just why the ability of those of ordinary skill in immunology to comprehend and perform the methodology described herein is questioned by the Examiner has been wholly unclear and remains so.

The reference, "Critical Synergy: The Biotechnology Industry and Intellectual Property Protection" pages 100-107, has *not* been shown by the Examiner to have any relation of any nature to this invention. The Office Action states that it "was geared to specific areas of technology, which applicant deems without any evidence to be non-inclusive of immunoassays, antigens and antibodies" (p.6). But the *Examiner* cited this reference; applicants did not, and it is the Examiner's responsibility to show specifically how it relates to the present invention. Having *read* this reference, consisting of 7 pages of a much larger document that is *unavailable* from any of the Patent and Trademark Office, the publishing entity (the Biotechnology Industry Organization) and a wide selection of the best library sources of scientific literature in the United States, and found *no mention* in the cited seven *pages* of "immunoassays", "antigens" or "antibodies", what is unreasonable about deeming it "noninclusive" of these *unmentioned* subjects? Especially so, when the subjects that *are* mentioned in this 7 page fragment, are quite different from, and unrelated to, the science of immunology which, compared to what the fragment deals with, is a mature technology? The Office Action's comment concerning the "Applicants' inability to obtain the entire document" (p.6) is believed to be an "inability" shared by the Examiner, moreover, since Applicants' counsel *did in fact* contact the Examiner for suggestions about locating the entire document and/or other related assistance, if possible, and was advised that *only the cited 7 pages* is available. *If* the Examiner sees some relevance to any part of this application in the cited 7 pages, she is courteously requested to point it out specifically; if not, the Examiner is requested to withdraw this alleged reference.

Parts of the Office Action appear to suggest that the disclosure is insufficient because *all* of what is necessary to obtaining highly antigen-specific antibodies to bacterial carbohydrate antigens, such as the methodology for separating selected carbohydrate antigens from the bacteria species, or serogroup, of which they are characteristic, is not highly novel--thus, the statement bridging pages 5 and 6 of the action that "There appears to be no conception of a method for detecting the presence of a carbohydrate antigen characteristic of at least one species or serogroup of a species of bacteria, i.e., both gram negative and gram positive bacteria using one purification procedure for any type of bacteria". Such a method, *if it existed*, would be highly novel. The fact that each bacteria species, or serogroup of a species, from which one may want to separate a target carbohydrate antigen in essentially protein-free form, based on work already done and reported in the literature, is known to need a purification/separation method differing somewhat from that for other bacteria species or serogroups and even a method differing from that for obtaining other different carbohydrate antigens characteristic of the same bacteria species or serogroup is what led to the various statements in the specification to the effect that *any* applicable method of antigen separation/purification may be used so long as the resulting antigen is essentially protein-free (i.e. contains less than 10% protein (wt/wt.).

In short, the Examiner correctly perceives that there is *no* generic purification procedure for obtaining a carbohydrate antigen from any type of bacteria disclosed; moreover, Applicants' position is that any effective purification method that yields a carbohydrate antigen having a protein content of not more than 10% may be utilized. Whether the antigen is characteristic of a gram-negative or gram-positive bacteria species, or serogroup, does play a

role, as the prior art teaches, in the details of the purification method that is selected--but once the carbohydrate antigen containing not more than 10% protein is obtained, there is a generic set of method steps that applies–i.e. the purified antigen is applied to a chromatographic affinity column; antibodies raised either to the bacteria from which the antigen was obtained or to the crude form of the same antigen are passed over it and the antibodies attain greatly enhanced antigen specificity with greatly improved ability to detect the crude form of the antigen, especially when that crude form is present in human (or mammalian) bodily fluids.

The assertion of nonenablement here states that " there is no purification guidance for different types of bacteria." (Office Action p.1) In fact, the application at page 14 clearly states that

> "Many methods for effecting these separation and purification steps are known in the literature and may be substituted for those herein described without departing from the scope of this invention, so long as the purified antigen obtained is essentially protein free as herein specified".

Furthermore, the technical literature includes a wealth of information on this subject which one skilled in immunochemistry can readily find and utilize, as the Pälin declaration submitted herewith points out.

The Examiner then says that "the specification at page 14 teaches different purification of carbohydrate antigen steps, including an incubation step, sonication step, repeated precipitation and centrifugation steps, lyophilization,...Lowry assay for proteins and tested for carbohydrate by phenol-sulfuric acid method." (Office Action p.6)

Of the steps recited in the quotation, however, *most* are not "purification" steps at all. Thus:

1) "incubation" apparently refers to the mixture of bacterial broth culture supernatant and cetyltrimethylammonium sulfate solution in a container that was "incubated" in an ice bath overnight--the first step in extracting the exemplified carbohydrate antigen from the bacterial culture broth. It could as well be said that the culture supernatant and the cetyltrimethylammonium sulfate solution were mixed and their container was placed in an ice bath and left overnight. There is no mystery about how to "incubate" in this sense. There is certainly nothing here that a person of ordinary skill in the art would fail to fully understand.

2) "sonication" is a step of concentrating (rather than purifying) a solution by aspirating off unnecessary liquid, usually water. "Sonication" *in fact*, is not mentioned on page 14 of this application, but does appear on page 15. It is a non-mysterious term, well understood by persons of ordinary skill in chemistry generally and immunology generally as well.

3) "repeated precipitation and centrifugation" are common techniques for effecting separation and purification steps. Their use is common and commonly understood by persons of ordinary skill in immunological and other chemical procedures.

4) "lyophilization" is likewise a common, well understood technique among those of ordinary skill in immunology, albeit *not* a purification step.

5) The Lowry assay for proteins and the carbohydrate test with phenol and sulfuric acid are common and conventional tests for protein content and carbohydrate content of a sample, respectively. *Neither* is a purification technique. Both are commonly utilized in verifying that a product recovered after several steps have been performed on a mixture *is* the product that was expected and both tests are routinely performed by and well understood by persons of ordinary skill in immunology and immunochemistry.

In point of fact, pages 14 and 15 of the application give a detailed description of an actual separation of a capsular carbohydrate antigen from *Haemophilus influenzae* type b. Just what is considered to be lacking about this description is wholly unclear. A specific example of an actual procedure is *all* that a patent applicant is required to provide.

The ensuing comment that "It is well known in the art that specific bacterial species require specific extraction methods, yet the claims do not take this into consideration and generically claim a method of detection" (O.A.p.6) seems *unrelated* to the previous sentence and what it intends to convey is exceedingly unclear. If this is important and "well known in the art", where is the reference or references that show it? What does it have to do with the ability of those of ordinary skill in the art to follow any of the known methods for separating carbohydrate antigens from bacteria, any of which may be utilized so long as the resulting antigen has a protein content not more than, and preferably less than 10% by weight? *Why* should the claims recite techniques or steps well known in the art that do not affect the outcome of any aspect of what the claims recite that is novel?

On the other hand, if the reference to "specific extraction method" in the sentence quoted in the preceding paragraph is intended to refer to the "extraction" of antigens from whole bacteria in samples, i.e., the opening of cell walls to make antigens more accessible to antibodies, referred to in Imrich et al patent 5,415,994, which patent covers a device having an "extraction chamber" for chemically treating analyte samples to break down bacterial cell walls and expose the antigen within them before conducting an assay, Applicants' experience and that of their assignee is that such "extraction" is *unnecessary* when the samples to be assayed are bodily fluid samples. See the Palin declaration ¶16(b), pp.10-11.

The specification, contrary to the sentence bridging pages 6 and 7 of the action includes:

1) a complete exemplification of how to obtain an essentially protein-free carbohydrate antigen from *Haemophilus influenzae* type b bacteria

2) a complete exemplification, incorporated by reference from copending Ser. No. 09/397,110 and its parent application Ser. No. 09/156,486 of how to obtain the essentially protein-free carbohydrate antigen that is found in all serogroups of *Streptococcus pneumoniae* and

3) a complete exemplification, (incorporated by reference from copending Ser. No. 09/139,720) of how to obtain the protein-free carbohydrate antigen of *Legionella pneumophila* serogroup 1 from a culture of *Legionella pneumophila* serogroup 1 bacteria.

In each instance, applicants have specifically exemplified a method of coupling the essentially protein-free bacterial antigen to a chromatographic column and a method for passing crude antibodies over the column and obtaining purified antibodies with high antigen-specificity for the unpurified, but otherwise identical bacterial antigen contained in a sample of human bodily fluid. Also described and exemplified in each instance is an ICT test strip for conducting an assay utilizing the purified antibodies to detect the same carbohydrate antigen of the bacterium in a sample of human bodily fluid from persons suspected of having a disease state attributable to the bacteria species, or serogroup of a species, from which the essentially protein-free antigen was extracted and against which the antibodies were raised.

Moreover, applicants' assignee has now had on the market for several years highly specific and sensitive assays for each of (1) the characteristic O-carbohydrate antigen of *Legionella pneumophila* serogroup 1 and (2) the carbohydrate antigen that occurs in and is characteristic of all serotypes of *S. pneumoniae*. See the Palin declaration ¶10(b), p.11. The number of these assays sold and successfully used, in the United States and other countries, is well into the millions and growing. These assays have been widely accepted as great boons to early detection and diagnosis of disease states caused by the corresponding bacteria.

On page 7, the office action appears to ignore that the specification defines the term "essentially protein-free", as used in this application, at p.6 of the specification as meaning antigen "containing not more than about 10% of protein". Again at page 13, second paragraph, the specification states

> "As used herein the expression "essentially protein-free state" means a state containing not more than-and preferably less than-about 10 percent (wt./wt.) of protein."

The action at p. 7, however, states that "The disclosure does not teach how to achieve the instantly claimed property or assurance of particular results which would be obtained if certain directions were pursued producing an essentially protein free carbohydrate bacterial antigen which is a highly empirical process." This sentence seems to state (insofar as it can be interpreted) that producing a bacterial carbohydrate antigen that contains less than 10% protein on a wt./wt. basis is a "highly empirical process". Applicants disagree. Example 2 at page 15 shows that the carbohydrate antigen of *Haemophilus influenzae* type b obtained contained 5% protein (wt./wt.) when analyzed by the Lowry assay for protein. Application Ser. No. 09/139,720, incorporated by reference in this application, shows that a carbohydrate antigen of *Legionella pneumophila* serogroup 1 was obtained (which is the same as that used to purify the antibodies utilized in the Binax NOW® test for detecting the crude form of the same antigen) which meets this criterion for "essentially protein-free", while application Ser. No. 397,110 also incorporated by reference, shows that the carbohydrate antigen of the *Streptococcus pneumoniae* species (which is the same as that used to purify the antibodies utilized in the Binax NOW test for detecting the crude form of that antigen) therein obtained also meets this criterion.

Summing up, the Examiner is *wrong* in characterizing as "highly empirical" Action (p.7) the methods which result in production of an "essentially protein-free carbohydrate bacterial antigen"--i.e. one that contains not more than 10% protein on a wt./wt. basis. The existing scientific literature is replete with descriptions of techniques for minimizing the protein content of carbohydrate antigens.

Applicants emphasize that the prior art, as the Palin declaration (¶¶7 and 8; see also ¶13) points out, dispels the Examiner's postulate of "unpredictability and complexity in the art" (Action, p.8) with respect to obtaining "an essentially protein-free bacterial carbohydrate antigen" (i.e. one containing not more than 10% by weight of protein). The Palin declaration itself likewise dispels the Examiner's "undue experimentation" (*Id.* p.8) premise by further pointing out that in addition to the wealth of literature available about methodology, suppliers of laboratory equipment and reagents furnish catalogs containing useful information about the performance capabilities of their products and how to use them. These suppliers also make available technical support personnel who are familiar with their products and area able to furnish valuable advice about selection of equipment and reagents for particular tasks.

In ¶16(e), p.14 the Palin declaration also emphasizes that Ser. No. 09/458,998's disclosure, referred to at p.8 of the action, of using a *different* "Reagent A", (which in this instance is a tris base containing SB3-8, a zwitterionic detergent) to "extract" and expose the target antigen present in *whole* bacteria that have been living in dormant or stagnant water, is *unrelated* to any assay conducted on mammalian bodily fluids and is concerned with opening cell walls and, in some instances, husklike walls further encasing them, of live bacteria growing in environmental water--a problem that is *not* manifested by the bacteria in human (or other mammalian) bodily fluids.

At page 8, the office action also endeavors to suggest that "Reagent A" comprising Tween 20, sodium azide and sodium dodecyl sulfate in sodium citrate phosphate buffer is an *essential* part of the claimed ICT assay, needed "to produce [i.e., extract] the crude carbohydrate antigen." As the Palin declaration points out in ¶¶16(b) and (c), pp. 10-13, this

is not correct. There is, in general *no need* to "extract" crude carbohydrate antigen contained in a human bodily fluid from the bacterium to which it is indigenous, because body processes unually have already done this (id, p.12). As ¶16(d) pp. 13-14 of Dr. Palin's declaration indicates the combination of buffer, surfactant and salts in "Reagent A" has a major function of augmenting sample volume to promote flow along the immunochromatographic strip and the further attribute that the surfactant inhibits non-specific binding during antibody-antigen reactions while the buffer and salts tend to provide a milieu that facilitates the antibody-antigen reactions. It is further pointed out that other common buffer-salt-surfactant solutions could also be used to perform the same functions.

The "Reagent A" mentioned in one of the specific examples of *this* application Serial No. 09/518,165 has *no* relationship to the "Reagent A" employed in Application No. 09/458;998, contrary to the assumption made on page 8 of the Office Action. As Dr. Palin's declaration points out,

(¶¶16(e) and 16(f) pp.14-15) Application 09/458,998 is concerned with an enzymeimmunoassay for an O-carbohydrate antigen of *Legionella* contained in bacteria colonizing in environmental water. Typically, the Legionella bacteria in such a milieu are intact with *unruptured* cell walls and often with a tough-husklike membrane covering them. To open the cell walls so that the affinity-purified antibodies disclosed in Application 09/139,720 can reach the target antigen requires the tris base/SB3-8 solution disclosed in Application Serial No. 09/458,988 or mechanical maceration, or both, as that application also discloses. As Dr. Palin's declaration points out, however, no such problem has been encountered in assaying for target carbohydrate antigens in human fluid samples.

Contrary to page 9 of the Office Action, Applicants have *not* made any "statements that specific sample types perform extraction on some bacteria". What Applicants *have* said is that the *Legionella* bacteria samples collected from environmental water often need their cell walls opened by mechanical and/or chemical means so that an enzymeimmunoassay can be performed on them. Applicants' relevant pending applications, apart from Serial No. 09/458,998, including Serial Nos. 09/139,720; 09/156,486 (now abandoned) and 09/397,110 make *no* reference to "extraction" of bacteria to open their carbohydrate antigens to antibody action because it is *not* needed when the bacteria are present in samples of human bodily fluid.

It is important, moreover, that Applicants have *never* identified any "sample types" that act to perform "extraction" on any bacteria. In Application 09/458,998, the "sample types"--environmental water samples containing growing bacteria--*are* themselves treated mechanically and/or chemically but these samples do not "perform extraction"--i.e. opening of cell walls. Applicants know of *no* "*sample* types" that have this function, in the context of the identified applications or otherwise.

Applicants also note that the claims of the present application, apart from claim___, are limited to assays performed on human bodily fluids and that it is their position that "extraction" of bacteria in those fluids to open up bacterial cell walls is *not* needed. Moreover, as Dr. Palin's declaration (p.13) points out, the reagents and methods needed to "extract" antigens from within bacterial cell walls and open them to antibody action are well known to immunologists and readily available in the literature and can always be included in any assay where experience shows they may be needed *without* in any way constituting an inventive step.

The rejection bridging pages 9 and 10 of the action is based upon an unclear premise and is believed to be wholly unjustified. By definition, the carbohydrate antigens of gram-positive bacteria belong to a class of compounds identified as "teichoic acids", lipoteichoic acids and derivatives of either". Just why applicants should somehow have endeavored to define these "derivatives" with specificity in this application is not clear. Applicants are *not seeking* to *make* derivatives of lipoteichoic or teichoic acids; the specification indeed in no sense suggests this. Moreover, it is axiomatic in immunology that antigens can be detected by antibodies and *vice versa* (i.e. antigens can be used to detect antibodies) but antigens cannot detect antigens. It is a fundamental principle of immunology that antigens and antibodies are the natural binding partners of one another. The only aspect of *derivatives* of teichoic or lipoteichoic acids that has any relevance to this application is that a carbohydrate antigen of an infectious bacteria species, or serogroup thereof, selected as a target antigen *may* be a derivative, in whole part, of either acid. In such a case, antibodies against the antigen can be raised in a conventional manner in a host animal by injecting the animal with the bacteria or with the crude antigen in solution, waiting for antibodies to develop in the animal over a time period of weeks or months and bleeding the animal, and separating the antibodies, which may then be affinity--purified as described in this application. It is not necessary, in order subsequently to employ these antibodies, to detect crude target antigen in a sample of human bodily fluid, to know the specifics of the functionality of either the antigen or the antibodies because an antigen and the antibodies raised against it or its host bacteria are, by definition in immunology, natural binding partners. The purpose and effect of the affinity purification described in this application is to *enhance* the natural binding proclivity of the antibodies for that antigen. Under these

circumstances, it is indeed difficult to comprehend the Examiner's position that the precise functionality of "derivatives" to teichoic or lipoteichoic acid that may exist as bacterial carbohydrate antigens in gram-positive bacteria needs to be defined and proved.

The Examiner's comments bridging pages 10-11 of the present action which are to effect that O-polycarbohydrate antigens are necessarily the only target antigens of *Legionella* bacteria that could be detected in the generally claimed process of this invention are likewise unfathomable. Applicants agree that Application Serial No. 09/139,720 discloses in detail working with an essentially protein-free O-carbohydrate antigen to affinity-purify antibodies and using those affinity-purified antibodies to detect the same crude carbohydrate antigen in human bodily fluids. *This* application Serial No. 09/518,165 , however, uses the detailed disclosure in that application, all of which is incorporated by reference herein, as an *example* within a generic concept applicable to the indigenous *carbohydrate antigens of bacteria in general*. Various *Legionella* bacteria species or serogroups of species are known to have other carbohydrate antigens that are within the scope of this general concept. Many bacteria species, including some of those of *Legionella* are further known to have multiple carbohydrate antigens. There is accordingly no reason why the claims here should be limited to O-linked-carbohydrate antigens. There can be no justification for the attempted limitation, in the absence of some showing of why the claimed methodology would not work with other carbohydrate antigens typical of Legionella species, or serogroups of species.

The Examiner's further comments in the paragraph bridging pages 11 and 12 of the Action are inapplicable to the present claims which no longer recite "esters" of teichoic and lipoteichoic acids.

The final full paragraph on page 12 of the Action prior to cumbered topic 7 rests upon a wrong premise which the application itself *and* the Palin declaration refute. Applicants in this generic application are *not* prescribing any specific carbohydrate antigen purification method. The disclosure tells the person of ordinary skill in the art, wishing to develop an assay for a carbohydrate antigen that is characteristic of a bacteria species, or serogroup, other than the carbohydrate antigens covered by the specific examples of this application (*including* those incorporated by reference) to utilize any known methodology for carbohydrate antigen separation/purification from bacterial culture so long as the antigen produced is an essentially protein-free embodiment as "protein-free" is herein defined. The Palin declaration attests that both the selection of purification separation methodology and the achievement of an essentially protein-free carbohydrate antigen embodiment are well within the ordinary skill in the field of immunology and were placed there by a wide variety of published literature. There is absolutely *no* need for *non-routine* experimentation here. It is noted that the specification points out in multiple places the essence of what is said above. See for example, p.14, lines 3-5, i.e.,

> "Many methods for effecting these separation [*in context*, of carbohydrate antigen from bacteria] and purification steps are known in the literature and may be substituted for those herein described, without departing from the scope of this invention, so long as the purified antigen obtained is essentially protein-free as herein specified".

VI. THE NEW MATTER REJECTION IN NUMBERED PARAGRAPH 7

Claims 37 and 46 have been amended to delete their recitation to "esters" of lipoteichoic and teichoic acids. Because the specification refers several times to the carbohydrate antigens of gram-positive bacteria as teichoic acids and lipoteichoic acids and their derivatives, specifically at p.2 in the first 4 lines after the "BACKGROUND OF THIS INVENTION" heading, on page 7 in lines 16-17, on page 9 in lines 5-6 under the heading and on page 12, lines 10-11, reference to "derivatives" has been subsituted for "esters" in those claims.

Applicants submit that since the term "derivatives" as applied to teichoic and lipoteichoic acids has always been present in the application, it cannot properly be characterized as "new matter". Attention is also invited to page 4 of the specification, lines 15-17 which state that the C-polysaccharide cell wall antigen of *Streptococcus pneumoniae* incorporated herein by reference form Ser. No. 09/397,110 "is a phosphocholine-containing polysaccharide derived from teichoic acid"--in other words, a *derivative* of teichoic acid. The same disclosure that this antigen is a derivative of teichoic acid also appears in Application Ser. No. 09/397,110 and its now abandoned parent application, Ser. No. 09/156,486. As already noted, the language herein denominating the carbohydrate antigens of gram-positive bacteria as teichoic and lipoteichoic acids and derivatives of either was taken from prior art descriptions and was not then and is not now believed to be controversial in any respect. It is further noted that since the teichoic and lipoteichoic acids and their derivatives *are themselves carbohydrate antigens*, they play no role whatever in detecting antigens. To the extent the action says that "one of

skill in the art would be required to perform undue experimentation to use esters of either lipoteichoic or teichoic acids to detect the crude antigen" (p. 12) it postulates an utter impossibility. Antigens cannot detect antigens; only antibodies can--and the present application in no sense discloses or suggests that the present invention involves using any derivative of teichoic or lipoteichoic acid for *detection* of a crude antigen. The application instead says that the carbohydrate antigens of gram-positive bacteria *are* teichoic or lipoteichoic acids and their derivatives, as the prior art in fact teaches.

VII. THE INDEFINITENESS REJECTION IN NUMBERED PARAGRAPHS 8, 9, 10 AND 11

The rejection of former claims 22-52, now in part replaced by new claims, as being unclear because they do not recite the origin of the antibodies is traversed. The specification at p. 13, lines 1-7 sets forth the preferred mode of raising antibodies--i.e., by injecting an animal such as a rabbit of goat with the crude target antigen or an IgG cut thereof. As those acquainted with immunology will recognize, the antibodies could also be raised against the bacteria to which the antigen is indigenous by injecting the animal with such bacteria preferably after first subjecting them to heat-killing. In fact, it is basic in immunology that there is no other way of obtaining polyclonal antibodies than injecting a mammal with the bacteria containing the antigen, which is the crudest and least refined antigen preparation that can be used or with crude antigen alone or with an IgG cut of the crude antigen and then bleeding the mammal after a suitable lapse of time to obtain serum containing antibodies.

Polyclonal antibodies are often purchased from laboratory businesses that specialize in raising them in animals maintained to serve as hosts for this purpose. All of this being well-known, Applicants do not understand the point of this rejection. Most patent claims involving polyclonal antibodies do *not* contain recitations about "*making* these antibodies"--in large part because the only way to "make" them is in a host animal. In an effort to provide the requested "clarification", language has been included in the present main claims to identify the antibodies passed over the column as raised in a mammalian host against the target antigen in crude form.

Step (d) of the claim that replaces claim 22 is intended to be generic to any type of assay in which the affinity-purified antibodies of this invention are employed to detect the crude target antigen in a liquid sample. Claim 53, which replaces claim 22, does include a step of contacting antibodies with the affinity purified antibodies and another *step of* detecting any antibody-antigen reaction product that is formed if target antigen is present in the sample. The word "counterparts", has been excised from replacement claim 53 in favor of language that specifies that the essentially protein-free antigen containing not more than 10% protein and the crude target antigen are both embodiments of the *same* target antigen. In claims 52, 53 and 54 language has been added to indicate that the moveably deposited antibodies in the first zone were labelled before they were so deposited.

Applicants traverse the rejection in ¶9 based on alleged indefiniteness of "derivative of either" for all of the reasons already stated herein relative to derivatives of teichoic and lipoteichoic acid that may be carbohydrate antigens of bacteria, noting that claim 24 has been cancelled.

As noted above, claims 36 and 45 no longer recite "esters" of teichoic acid and lipoteichoic acid, but have been revised to recite "derivatives" of these acids consonant with the specification.

Claims 35-36 and 44-45 have been amended to correct the errors pointed out in ¶11 of the Action.

VIII. INCORPORATION BY REFERENCE--NUMBERED PARAGRAPH 12, OF THE ACTION

As previously pointed out on several occasions, the position taken in this action regarding incorporation by reference is in direct conflict with M.P.E.P. 608.01(p) A1 which *encourages* incorporation by reference of United States copending, commonly assigned patent applications.

Each of the cited decisions *In re Hawkins,* 179 USPQ 157 (CCPA 1973); *In re Hawkins* 179 USPQ 163 (CCPA 1973) and *In re Hawkins*, 179 USPQ 167 (CCPA 1973) is concerned solely with attempted incorporation by reference of British --i.e., *foreign*--patent applications, as to which MPEP Section 608.01 (p) A2 states incorporation by reference may not be used.

Applicants have requested in the past and here repeat the request that the Examiner please *explain* why a premise concededly applicable to foreign applications is being applied to U.S. patent applications in derogation of the MPEP. The mere assertion that Applicants are attempting to incorporate essential material by reference, which is then said to be "improper", is *not* at all *consistent* with what is said in either M.P.E.P. section 608.01 (p) A1 or MPEP section 608.01 (p) A2.

The sense of MPEP 608.01 (p) A1 is that *any* material one chooses, whether or not essential material, that is already disclosed in a U.S. patent or patent application *not only can, but should* be incorporated by reference in a later application. The sense of MPEP 608.01 (p) A2 is that material disclosed in a foreign application should not be incorporated by reference, but should be repeated if desired to be incorporated in a U.S. application. There is *nothing*

in the *Hawkins* decisions that forecloses incorporation by reference in a U.S. application of essential material contained in an earlier filed copending U.S. application.

IX. "DOUBLE PATENTING" ¶13 OF THE ACTION

Applicants traverse the rejection based on alleged conflict between claims of this application and certain claims of application 09/458,998.

As the claims herein have been amended herewith, Claim 53 and its dependent claims are of broader scope than any claim of Application 09/458,998, while there is a clear line of demarcation between those claims herein that cover conducting an ICT assay for crude target antigen present in a human bodily fluid and *all* of the claims in Application Serial No. 09/458,998, all of which are limited to conducting an enzymeimmunoassay for target antigen in colonized bacteria having intact cell walls that are growing in environmental water which, if obtained from heating/air conditioning systems, may contain e.g., inorganic materials such as rust and other residues from old pipes and other heating or cooling devices and if obtained from stagnant outdoor ponds or pools, may contain molds, mosses, and other non-human residue.

To the extent that any of the claims of this application overlaps any claim of any of the Applications 09/139,720; 09/458,998 or 09/397,110 that have been incorporated herein by reference, the common assignee of these applications is prepared to submit an appropriate terminal disclaimer or disclaimers at such time as it becomes clear in which of these applications such a disclaimer is appropriate. Since that time has not yet arrived, no more can presently be done.

X. THE 35 U.S.C.§103(a) REJECTION-PARAGRAPH 14 OF THE ACTION

The rejection is improper because it rests upon a combination of references which, even if, *arguendo,* capable of producing a similar end result (which Applicants seriously doubt that it is) does not when combined, achieve the invention herein disclosed and claimed. The Examiner relies upon a monoclonal antibody disclosed by Barthe *et al* which is said in the action to recognize several "several crude carbohydrate antigens from Legionella" albeit this is not so clear in the article. Moreover, the object here is an assay in which the antibodies consistently recognize *one* and only one target antigen on a reliable and consistent basis.

There is nothing in the Barthe article or in what the Examiner says, moreover, that suggest this monoclonal antibody would be even remotely useful in a rapid ICT test, or that it would recognize a target antigen in a bodily fluid sample in a rapid test. The test described by Barthe are all of an obviously time-consuming nature requiring dedicated attention of highly skilled, trained laboratory workers including an ELISA test, an indirect immunoflorenscence test, and a Western Blot test.

Moreover, there is no indication that this Barthe alleged "monoclonal" antibody was specific to a particular antigen characteristic of either a bacteria species, or a serogroup of a bacteria species, so that identification of the antigen by the antibody necessarily confirms the presence of its host bacteria species, or serogroup of a species, known to be causative of a particular disease in bodily fluids of a human patient.

While the Examiner is correct that limitations and other characteristics of an invention not recited in the claims usually cannot be relied on to distinguish a reference, the Examiner in this application has to read multiple limitations *out* of the claims and ignore them, to rely on this attempted somewhat ungainly combination of Imrich and Barthe *et al.* It is further noted in this regard that disclosed *uses* of an invention, even if not claimed, frequently *are* properly availed of to distinguish prior art.

The invention in this case is *not* directed to making or using monoclonal antibodies which, by definition are normally obtained by the published method of Kohler and Milstein from mouse hybridomas, as describe by Barthe *et al* on page 1016, right hand column of their article.

It is directed *instead* to a particular affinity purification of polyclonal antibodies wherein the purifying agent on the affinity column is an essentially protein-free carbohydrate antigen (where "essentially protein-free means" not more than 10% (wt./wt.) of protein). Even if it were possible to show that the Barthe monoclonal antibody used in the Imrich device were able to achieve essentially the same test result in a test to detect an antigen characteristic of *Legionella pneumophila* serogroup 1 in patient urine samples as the result that Applicant's affinity-purified antibodies produce in applicant's ICT device and method when, e.g., patient urine samples are tested, this would not make applicant's novel affinity purified polyclonal antibodies or their use in an ICT assay obvious in relation to the *different* Barthe antibody as used in the Imrich test.

The Examiner overlooks completely that a §103(a) rejection can only be appropriate if the reference combination renders the "subject matter as a whole" of the invention under discussion obvious to one of ordinary skill in the art. Applicant's affinity-purified antibodies are novel and their use in rapid tests that permit early diagnoses of diseases that, prior to the introduction of these tests, required bacterial cultures and several days delay before diagnoses could be made, are in no sense "obvious" from an attempted combination of Imrich and Barthe.

The Imrich-Barthe *et al* reference combination of th action altogether ignores the requirements of *all* the claims that crude polyclonal antibodies or an Ig G cut thereof be passed over an affinity column to which is coupled an embodiment of the target antigen containing no more (and preferably less) than 10% of protein and moreover, that the antibodies so purified be used to detect the target antigen in samples. Those claims that are limited to the preferred ICT test further require that the ICT strip be prepared by placing a movable deposit of the antibodies in labelled form near the sample introduction point and an *immovable* deposit of unlabelled antibodies at the capture line near the opposite end of the strip, and further require that the sample to be tested by human body fluid *and* that the test result be available within 15 minutes of the introduction of sample to the strip. The Imrich reference does not even suggest any of these requirements except the use of a sample from a human patient. The Barthe *et al* article contains no disclosure of *any* of these requirements. Without what is lacking in the reference combination, there is *no way* for "the subject matter as a whole" to be rendered obvious even to a person of the highest possible level of skill in immunology, and much less to the person of ordinary skill addressed in the statute.

It is also noted that the Office Action appears to expect exceedingly low levels of skill from the person of ordinary skill, far below those that actually exist, when addressing the issues of enablement raised in this action. Yet when addressing the issue of obviousness to one of ordinary skill in the art as posited upon the Imrich-Barthe *et al* reference combination, the person of ordinary skill in the art suddenly is envisioned as having extraordinary skill and a virtually extrasensory perception that allows him or her to leap from a description of the proverbial sow's ear to a clear conception of a silk purse.

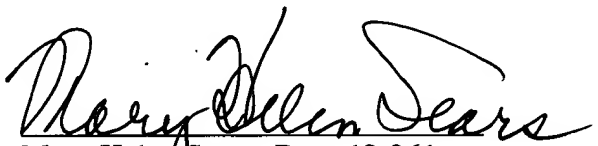
The 35 U.S.C. §103(a) rejection is flawed and should be withdrawn.

CONCLUSION

Applicants believe this application is in condition for allowance and accordingly respectfully request such action.

Should the present rejections nonetheless be substantially adhered to, Applications request that the next action be made final, since it appears in that event that an impasse has clearly been reached and that any further pursuit of this application should proceed in an appeal.

Respectfully submitted,



Mary Helen Sears, Reg. 19,961
Attorney for Applicants
The M.H. Sears Law Firm, Chartered
910 Seventeenth Street, N.W., Suite 800
Washington, D.C. 20006
Telephone: (202) 463-3892
Telecopy: (202) 463-4852